REMARKS

Status

In the Office Action mailed May 6, 2003, Applicants canceled Claims 1-20 and added Claims 21-34.

Presently, Applicants cancel Claims 26 and 27 in favor of new Claims 35 and 36, which are directed to the invention described as Group I by the Examiner, which is drawn to an immunogenic composition comprising a viral antigen and ribavirin, wherein the viral antigen is a nucleic acid. Support for Claims 35 and 36 can be found throughout the specification (*e.g.*, page 4, lines 4-8, page 5, lines 11-14, and page 8, lines 15-18). Applicants also amend Claim 29 to require that the nucleic acid component comprises a fragment of the sequence of SEQ. ID NO. 13 that encodes an antigenic peptide. Support for this amendment can also be found throughout the specification (e.g., page 26, lines 8-15).

Response to Restriction Requirement

The Examiner has requested that Applicants restrict prosecution to one of two groups of claims:

I: Claims 21-25 and 28-34 drawn to an immunogenic composition comprising a viral antigen and ribavirin, wherein the viral antigen is a nucleic acid molecule; and

II: Claims 26-27 drawn to an immunogenic composition comprising a viral antigen and ribavirin, wherein the viral antigen is an amino acid molecule.

Applicants have elected to prosecute claims directed to the subject matter of Group I, immunogenic compositions comprising a viral antigen and ribavirin, wherein the viral antigen is a nucleic acid. Applicants confirm that in a telephone conversation with the Examiner on July 24, 2003, the election was made to prosecute the invention of Group I.

New Claims 35 and 36 are directed to the subject matter of Group I and consideration on the merits is requested.

Claims Objections

The Examiner has objected to Claims 21 and 30-34 because:

> The recitation of a 'nucleic acid' should be changed as 'a nucleic acid sequence or molecule' because an antigen cannot be made by a single nucleic acid. An antigen in a nucleic acid form is usually presented as a plasmid DNA, wherein the antigen is encoded [*sic*] at least [*sic*] couple of nucleic acids or a nucleic acid sequence. Therefore, a proper correction is requested. *(See page 3, ¶ 8).*

Applicants respectfully submit that the Examiner has confused the term "nucleotide" with "nucleic acid." One of skill in the art readily appreciates that a nucleic acid is comprised of several nucleotides and that a single nucleic acid can encode a peptide, which is an antigen. Accordingly, Applicants submit that because a nucleic acid is well understood to encode an antigen, the Examiner's objections to Claims 21 and 30-34 should be withdrawn.

Double Patenting

The Examiner has rejected Claims 21-25 and 28-34, provisionally, for obvious-type double patenting over the claims to co-pending U.S. App. No. 10/104,966. To overcome the provisional rejection of non-statutory double patenting, Applicants submit herewith a Terminal Disclaimer.

Rejections Under 35 U.S.C. § 112

The Examiner argues that Claims 30-34 are rejected under 35 U.S.C. § 112, ¶ 2, as being incomplete for omitting essential elements. The Examiner states:

> The omitted elements are: the concentration for the nucleic acid antigen and concentration for the ribavirin and how they are provided in the composition, i.e. the ratio of HCV nucleic acid antigen to the ribavirin etc. *(See page 4, ¶ 13).*

As an initial point, Applicants wish to focus the Examiner's attention to the fact that Claims 30-34 recite methods of making the claimed compositions.

Applicants also want to bring the Examiner's attention to the fact that the use of ribavirin is well known in the art. Tam (U.S. Patent No. 5,767,097A), for example, states:

> Since ribavirin has been on the market for several years, many dosage forms and routes of administration are known, and all appropriate dosage forms and routes of administration may be utilized. For example, in addition to oral administration, ribavirin may be given intravenously, intramuscularly, intraperitoneally, topically and the like, all of which are known. (*See* column 4, lines 25-41).

Applicants further want to point out that the use of nucleic acids as antigens in DNA-based vaccination is also well known. (*See, e.g.*, U.S. Patent Nos. 5,589,466; 5,846,946; 6,214,804; and 6,586,409). Enke et al., for example, report obtaining specific antibody responses to nucleic acid antigens encoding three different HCV non structural proteins and the authors also reference five other papers that report the use of genetic immunization *in vivo* to induce immune responses to pathogens such as influenza virus, HIV-1, and tuberculosis. (*See page 4921, ¶ 2 and references cited therein).*

Lastly, Applicants want to emphasize that formulation of immunogenic compositions with and without adjuvants is routine. U.S. Patent No. 6,586,409 for example, describes in detail the formulation of immunogenic compositions containing adjuvants for the purpose of polynucleotide-based vaccination.

As to the Examiner's arguments that amounts of ribavirin, antigen and ratios etc. are essential elements, Applicants point out that the specification is replete with evidence that ribavirin can be used as an adjuvant over a wide range of concentrations with varying amounts of viral antigen.

The specification states for example:

> The vaccines contain approximately 0.25 mg - 2,000 mg of ribavirin. That is, some embodiments have approximately 250 μg, 500 μg, 1 mg, 25 mg, 50 mg, 100 mg, 150 mg, 200 mg, 250 mg, 300 mg, 350 mg, 400 mg, 450 mg, 500 mg, 550 mg, 600 mg, 650 mg, 700 mg, 750 mg, 800 mg, 850 mg, 900 mg, 1 g, 1.1 g, 1.2 g, 1.3 g, 1.4 g, 1.5 g, 1.6 g, 1.7 g, 1.8 g, 1.9 g, and 2 g of ribavirin.

> Conventional vaccine preparations can be modified by adding an amount of ribavirin that is sufficient to enhance an immune response to the antigen. That is, existing conventional vaccine formulations can be modified by simply adding ribavirin to the preparation or by administering the conventional vaccine in conjunction with ribavirin (*e.g.*, shortly before or after providing the antigen). As one with skill in the art will appreciate, the amount of antigens in the vaccine can vary depending upon the antigen and its immunogenicity. The amount of antigens in the vaccines can vary accordingly. Nevertheless, as a general guide, the vaccines can have approximately 0.25 mg - 5 mg, 5 - 10 mg, 10 - 100 mg, 100 - 500 mg, and upwards of 2,000 mg of an antigen (*e.g.*, a hepatitis viral antigen). (*See* page 28, lines 21-31, and page 29, lines 1-5).

Furthermore, Example 1 shows that adjuvant activity was observed when ribavirin (1 mg, 3 mg, or 10 mg) was co-administered with two different concentrations of antigen. Example 2 shows that even low doses (less than 1 mg) of ribavirin were found to enhance an immune response to a co-administered antigen. (*See* Figure 2). Table 7 also shows that 0.1 - 10 mg of ribavirin was sufficient to enhance an immune response to a co-administered antigen. Clearly, the data in the specification support the finding that a wide range of concentrations of ribavirin can be used as an adjuvant when co-administered with various concentrations of antigen.

The specification not only describes immunogenic compositions containing various concentrations of ribavirin and various concentrations of nucleic acid that encode an antigen but it also describes various approaches to formulate an immunogenic composition comprising ribavirin and a nucleic acid. (*See* page 67, line 18 – page 69, line 13).

Applicants submit that the formulation of immunogenic compositions is well within the skill in the art and specific guidance is provided in the specification to manufacture the claimed immunogenic compositions according to Claims 30-34. Specific amounts, ratios, and formulations are not required to practice the invention nor should they be required limitations to Claims 30-34. Applicants request that the Examiner withdraw these grounds for rejection.

The Examiner also rejects Claims 21 and 29 under 35 U.S.C. § 112, ¶1, arguing that the specification does not reasonably provide enablement for a composition made by any piece of nucleic acid viral antigen, especially a whole HCV virus genome nucleic acid sequence in combination with ribavirin.

Applicants have amended Claim 29 to require that the immunogenic composition comprises a fragment of the sequence of SEQ. ID NO. 13 that encodes an antigenic peptide. Applicants agree, as the Examiner has pointed out:

> State of art*[sic]* teaches that the DNA vaccine can be made by injecting a plasmid into a host and a plasmid vector encoding HCV antigenic protein but not the whole HCV genome is able to induce an immune response after administering into an animal. However, it is unpredictable whether *[sic]* you inject *[sic]* whole coding sequence of HCV genome into a subject will induce an enhanced immune response or produce a replicating or infectious hepatitis C viral RNA since transfecting a subgenomic HCV into a cell line can produce infectious HCV RNA in vitro as evidenced by Lohman et al. (Science 1999, Vol. 285, pp. 110-113, see abstract) or an acute or persistent infection in vivo as evidenced by Forns et al. (PNAS 2000, Vol. 97, pp. 13318-13323, see abstract). (*See page 5,* ¶ 16).

Applicants point out that the preamble of Claims 21 and amended Claim 29 require, however, that the claimed compositions are "immunogenic compositions." As such, nucleic acid antigens that fail to elicit an immune response are not encompassed by the claims. Applicants also wish to emphasize that, based on the understanding in the field, as exemplified by the teachings above, one of skill in the art would readily appreciate that the use of the whole HCV genome would be inoperable in an immunogenic composition and, thus, one of skill in the art would not use the whole coding sequence of the HCV genome in an immunogenic composition. According to *M.P.E.P. §2164.08(b):*

> The presence of inoperative embodiments within the scope of the claim does not necessarily render a claim non-enabled. The standard is whether a skilled person could determine which embodiments that were conceived, but not yet made, would be inoperative or operative with expenditure of no more effort than is normally required in the art. *See M.P.E.P. §2164.08(b).*

Applicants have provided considerable data demonstrating that ribavirin is an adjuvant when co-administered with an antigen. The determination of whether a particular formulation of ribavirin and nucleic acid antigen enhances an immune response is straightforward and routine in

the field. It does not require undue experimentation to make and use the claimed compositions. The preparation and administration of DNA vaccines with and without adjuvants has been routinely practiced for several years and the determination of whether co-administration of a known antigen and ribavirin enhances an immune response requires only the step of comparing the immune response produced when the formulation includes ribavirin to the immune response produced when the formulation does not contain ribavirin. These experiments are described in detail in the specification. It does not require undue experimentation to perform these tests. On the contrary, these tests are routinely conducted during the formulation of any immunogenic composition containing an adjuvant. Applicants respectfully request that the rejections under 35 U.S.C. §112 be withdrawn.

Rejections Under 35 U.S.C. § 103

The Examiner has rejected Claims 21-25 and 30-34 under 35 U.S.C. § 103(a) as being unpatentable over Encke et al. (*Intervirology* 1999, 42:117-124), Tam, R. (U.S. Patent No. 5,767,097A) and Hultgren et al. (*J. Gene. Virol.*, 1998, 79:2381-2391).

The Examiner states that Encke et al. teach that DNA-based immunization of HCV antigens gave detectable antibody responses in subjects but that the authors do not teach the use of ribavirin in combination with the HCV antigens to produce an enhanced immune response.

The Examiner also argues:

> Tam R. teaches that *[sic]* method for produce *[sic]* an immune response, preferentially an enhanced TH1 type immune response to a specific antigen by administering a composition comprising a viral component with a non-viral component of rebavirn *[sic]* into patients (Claims 1-9). (*See* page 7, ¶ 21).

The Examiner then argues that Hultgren et al. teach a method for inducing an enhanced immune response by administering HBV and HCV antigens on the basis of daily administration of ribavirin. The Examiner states:

> Hultgren et al. teach a method for inducing an enhanced immune response by administering hepatitis viral antigen including HBV e antigen, core antigen and

> HCV core or non-structural protein NS3 on the basis of daily administering ribavirin 0-1.5mg/day in mice (See Methods on pages 2382-2383). They demonstrated that the injection of the composition is able to produce an enhanced immune response by increasing the anti-HCV NS3 or anti-HBe antibody production or TH1 type of cytokine secretion such as IL2 or INF-γ. (See entire section of results especially Figs. 3-5). *(See pages 7 and 8* ¶ 22).

Lastly, the Examiner argues that, with respect to the limitation of co-administration, regardless of whether the antigen and ribavirin are co-administered or are administered separately, the results are the same and, thus, the claimed invention is obvious absent unexpected results.

At the outset, Applicants want to emphasize that the term adjuvant, as it has been and is being used in the vaccine field, stems from the latin word "adjuvare" which means to help. Thus, an adjuvant should help, neither shift nor modulate but simply help, the antigen in the vaccine to become more immunogenic. Applicants have discovered that ribavirin can be used as an adjuvant (i.e., to enhance an immune response).

Although Encke et al. teaches that intramuscular DNA-based immunizations for various HCV antigens produce detectable antibody responses, as the Examiner has recognized, Encke et al. does not teach the use of ribavirin in combination with these antigens. The Examiner first tries to fill this gap with Tam by arguing that Tam teaches administration of ribavirin and a composition comprising a viral component. The Examiner, however, has misread Tam, which states in Claim 6:

> 6. A method of treating a patient having a disease which includes a viral component and a non-viral component, the non-viral component being characterized by reduced TH1 levels and increased TH2 levels in activated T lymphocytes, comprising administering ribavirin to the patient under a protocol sufficient to promote the TH1 response and suppress the TH2 response in a patient.

The claim recites the treatment of a patient with a viral disease ("having a disease which includes a viral component") by providing ribavirin. In Tam, there is no evidence, indication, suggestion or motivation for administering any composition with an antigen, much less a

composition comprising an antigen and ribavirin. Tam only describes that the daily administration of ribavirin results in immune -modulation.

Furthermore, Tam illustrates how the field teaches away from the claimed invention. Tam states:

> In addition, we have significantly advanced the prior research by demonstrating that ribavirin modulates the cytokine pattern of an immune response at least in part by promoting a Th1 response and suppressing a Th2 response. In hindsight, this discovery is not inconsistent with prior research. First, ribavirin is known to inhibit both functional humoral immune responses, (Peavy et al, 1981, J Immunol 126: 861-864, Powers et al, 1982, Antimicrob Agents Chemother 22: 108-114) and IgE-mediated modulation of mast cell secretion (Marquardt et al, 1987, J Pharmacol Exp Therapeutics 240: 145-149, (both Th2 lymphokine-mediated events). (*See Column 2, lines 65-67 and Column 3, lines 1-9).*

Thus, Tam teaches that ribavirin inhibits immune responses. There is, therefore, no suggestion or motivation to use ribavirin to enhance an immune response to an antigen.

The Examiner has also misread Hultgren et al. The Examiner refers to Figures 3-5 for the proposition that daily administration of ribavirin followed by periodic administration of an antigen enhances an immune response. In fact, the results of Hultgren are the exact opposite.

Hultgren et al., disclose that daily ribavirin therapy accompanied by an immunization with an antigen and Freund's adjuvant results in a slight shift in IgG subclass distribution, with the most marked increases in IgG2a and IgG2b, however, no differences were seen in the total IgG levels between the treatment groups. (*See Figure 4 and the discussion on page 2386, first paragraph).* This is immune-modulation not an enhanced immune response or adjuvant activity. Although Hultgren et al. describe the daily administration of ribavirin and periodic immunization with an antigen, their approach was different than Applicants co-administration protocol and the results obtained are significantly different. Applicants observed a rise in antibody titers to the specific antigen when ribavirin was co-administered with an antigen, whereas, Hultgren et al. saw no change in antibody titers, only a shift in IgG subclass distribution.

In fact, Hultgren et al., teach away from co-administration of ribavirin and an antigen to enhance an immune response to the antigen. The authors report, for example, that "[t]he highest dose of daily ribavirin completely prevented anti-HBe seroconversion whereas lower ribavirin doses reduced antibody titres (Fig. 4c)." (*See page 2387, first paragraph).* The authors also state that "[w]e show that ribavirin treatment causes a transient drop in HCV-specific humoral responses during treatment of patients with chronic HCV infection." (*See page 2388, last two lines and page 2389, first paragraph).* Still further, the authors state that "[r]ecent studies have indeed shown that ribavirin is immune-suppressive *in vitro*" and that "similar effects may well be present *in vivo* during treatment of chronic viral hepatitis." (*See page 2389, second paragraph).* Lastly, the authors report that "[h]igh daily doses of ribavirin (>1mg/day) applied to HBeAg-Tg mice totally inhibited anti-HBe seroconversion, clearly showing the immune-suppressive effects of ribavirin *in vivo*." (*See page 2389, fifth paragraph).*

With regard to the point that co-administration is the same as daily therapy followed by periodic administration of an antigen, Applicants submit that the results are not the same, as exemplified in Hultgren et al. Again, according to Hultgren et al., there was no increase in total antibody titer specific for the antigen when ribavirin was given daily and the antigen was administered periodically, whereas by Applicants co-administration protocol a rise in antibody titer was observed.

In contrast to the findings provided by Hultgren et al., Applicant's specification shows that when antigen and ribavirin are co-administered an enhanced immune response is obtained. (*See Figures 1, 2, and Table 1*). Figure 1, for example, shows that 10μg of antigen co-administered with 1 mg of ribavirin generates nearly the same mean antibody titer against the antigen as an immunization with 100 μg of antigen without co-administration of ribavirin. Figure 2 shows that wide ranges of ribavirin, when co-administered with antigen, produce this adjuvant effect. Table 1 shows that by adding ribavirin to a sub-optimal vaccine dose, a commercially available preparation, antigen-specific antibodies became detectable., however, in the absence of ribavirin, no detectable antibodies were observed. Clearly, the enhanced immune response obtained from co-administration of ribavirin and an antigen is significantly different and

unexpected from the immune-modulation obtained from daily ribavirin therapy combined with administration of antigen. Furthermore, Applicants state:

> As shown in Table 8, the addition of ribavirin to the immunogen prior to the injection does not change the IgG subclass response in the NS3-specific immune response. Thus, the adjuvant effect of a vaccine composition comprising ribavirin and an antigen cannot be explained by a shift in the TH1/TH2-balance. It appears that another mechanism may be responsible for the adjuvant effect of ribavirin. (*See* page 19, lines 5-10).

In contrast to the Examiner's assertion, the data support the conclusion that co-administration is **not** the same as daily therapy followed by periodic administration of an antigen. Co-administration of ribavirin and an antigen enhances an immune response, whereas, daily ribavirin therapy followed by periodic immunization with an antigen promotes immune-modulation. When ribavirin is co-administered with an antigen it produces an adjuvant effect but when it is provided as a daily therapy and an antigen is periodically administered a shift in IgG subclass is observed but no increase in total antibody titer specific for the antigen is obtained. Accordingly, co administration is not a limitation that can be ignored.

In sum, the Examiner has not made a prima facie case of obviousness because all of the limitations of the claims are not present or suggested by the prior art. Enke et al. teaches that DNA-based vaccination can be performed with various HCV constructs but there is no evidence or suggestion in this reference to co-administer these nucleic acid antigens with ribavirin. Tam teaches that daily ribavirin therapy produces immune-modulation but there is no evidence or suggestion in this reference to combine the daily ribavirin therapy with immunization of an antigen to increase the immune response. The closest reference, Hultgren et al., describes daily ribavirin therapy with periodic administration of an antigen. There is no evidence or suggestion in this reference that the ribavirin and antigen are co-administered and daily administration of ribavirin followed by periodic immunization produces immune-modulation not an enhanced immune response. Accordingly, it is not possible to argue that co-administered ribavirin and antigen and daily administration of ribavirin followed by periodic immunization produce the same results. In fact, the results Applicant obtained are unexpected because an increase in total antibody titer specific for the antigen was observed when ribavirin and an antigen were co-administered; whereas, the daily ribavirin and periodic introduction of antigen-type protocol, as

exemplified in Hultgren et al., did not produce an increase in total antibody titer specific for the antigen (*see Figure 4c in Hultgren et al.)* but rather produced a shift in IgG subclass (i.e., immune-modulation).

Information Disclosure Statement

Applicants submit herewith an Information Disclosure Statement, which includes an abstract that was cited by the Examiner in a related application (U.S. Pat. App. No. 10/104,966). Chiang et al., *Vaccine Strategies Against Microbial Pathogens*, April 2000, 42.14, is a pre-publication abstract that adds nothing to Hultgren et al. Chiang et al. disclose that daily ribavirin therapy combined with bi-weekly immunization of antigen increases the presence of IgG2a. Hultgren reports the same thing. (*See Hultgren et al., pages 2386, first paragraph* -- "small differences were seen in the IgG subclass distribution of the ribavirin-treated mice with the most marked increases in IgG2a and IgG2b (Fig. 4d-e)." As reported by Hultgren et al., daily ribavirin therapy combined with immunization with an antigen produces a shift in the distribution of IgG subclass (immune-modulation) but no change in the mean antibody titre specific for the antigen. The prior art is replete with references that report that daily ribavirin therapy is immune-suppressive and one of skill in the art would appreciate upon reading Chiang et al. that the authors were simply reporting that a shift in IgG subclass (immune-modulation) was observed. Accordingly, Chiang et al., does not teach or suggest Applicants' claimed invention, alone or in combination with any of the cited references. Applicants respectfully request consideration of the references cited in this Information Disclosure Statement on their merits.

Conclusion

The undersigned has made a good-faith effort to respond to the Office Action and to place the claims in condition for allowance and such action is earnestly solicited. Nevertheless, if any undeveloped issues remain or if any issues require clarification, the Examiner is respectfully requested to call Applicants' attorney, Eric S. Furman, Ph.D., at (619) 687-8643 (direct line) to resolve such issues promptly. Please charge any additional fees, including any fees for additional extension of time, or credit overpayment to Deposit Account No. 11-1410.

Respectfully submitted,

KNOBBE, MARTENS, OLSON & BEAR, LLP

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